

WHAT IS CLAIMED IS:

1. A method for analysis of proteins in a biological system comprising:

providing a biological system;

exposing the system to a stimulus;

5 sampling the biological system at multiple time intervals after exposing the system to the stimulus,

treating the multiple samples by separation technique to provide multiple protein samples suitable for analysis by mass spectrometry, and

10 analyzing the multiple samples to determine changes in protein abundance as a function of time after exposing the biological system to stimulus, said analyzing including providing a parallel array of mass spectrometry systems adapted for protein analysis, and

directing mass spectral data from the mass spectrometry systems in said array to a common computing device, said mass spectral data being indicative of the identity and the abundance of protein in said multiple sample, and

15 correlating said mass spectral data as a function of time.

2. The method of claim 1 comprising displaying said correlated data as a function of protein identity, protein abundance, and time.

3. The method of claim 1 wherein the correlated data is stored in a searchable database.

4. The method of claim 1 comprising identifying proteins based on changes in abundance as a function of time.

5. The method of claim 4 wherein said array includes at least 20 mass spectrometers.

25 6. The method of claim 4 comprising analyzing 500 proteins or more.

7. The method of claim 6 comprising analyzing 5000 proteins or more.

8. The method of claim 4 wherein the separation technique includes separation apparatus and said common computing device communicates with said separation apparatus.

9. The method of claim 8 wherein the separation technique includes chromatography.

10. The method of claim 8 wherein the separation technique includes use of a magnetic particle separation apparatus.

11. The method of claim 10 where the magnetic particle separation apparatus treats multiple samples in parallel.

12. The method of claim 4 wherein said mass spectral data includes peptide fragment mass spectra and an amino acid sequence derived from a data base.

13. The method of claim 12 wherein said mass spectrometer are LC-TMS mass spectrometers.

14. The method of claim 4 comprising exposing a first component of the biological system to a stimulus and maintaining a second component of the biological system free of the stimulus, sampling and analyzing each of the first component and the second component and comparing the identity and abundance in the first component and the second component.

15. The method of claim 14 comprising separately analyzing samples from said first component and second component.

16. The method of claim 4 wherein the stimulus is a drug.

17. The method of claim 4 wherein the time interval is about 5 to 60 seconds.

18. The method of claim 4 wherein the time interval is about one minute to one hour.

19. A system for mass spectrometric analysis comprising:

a parallel sample separation apparatus adapted to separate multiple samples in parallel
5 for analysis by mass spectrometry, and

a parallel array of mass spectrometry systems adapted to receive the samples from the separation apparatus, and

a common computing device communicating with the parallel array of mass spectrometry systems and the parallel separation apparatus, the common computing device
10 being adapted to analyze mass spectral data from the parallel array of mass spectrometry systems as function of sample identity.

20. The system of claim 19 where the parallel separation device is a parallel magnetic particle separation device.

21. The system of claim 19 wherein said array includes at least 2 mass spectrometers.
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22. A method for analysis of proteins in a biological system comprising:

providing a biological system containing proteins;

exposing the biological system to a stimulus;

after exposing the biological system to the stimulus, sampling the biological system at
20 multiple time intervals to obtain multiple samples;

treating the multiple samples by a separation technique to provide multiple protein samples suitable for analysis by mass spectrometry;

providing a parallel array of mass spectrometer systems capable of simultaneous analysis of as many protein samples as there are spectrometer systems in said array;

analyzing the multiple protein samples in said parallel array of mass spectrometry
25 systems to generate mass spectral data indicative of the identity and the abundance of proteins in said multiple protein samples; and

in a common electronic computing device communicating with each of said mass spectrometry systems, correlating said mass spectral data as a function of time.

23. The system of claim 22 where the parallel separation device is a parallel
5 magnetic particle separation device.

24. The system of claim 23 wherein the parallel array includes an array of LC-MS spectrometer system.

25. The system of claim 24 wherein the array includes 6-20 mass spectrometers.

26. The system of claim 25 wherein the time intervals are in the range of 5
10 seconds to 10 minutes.

27. The system of claim 26 wherein the analysis includes analysis of about 500 proteins or more.

28. The method of claim 27 wherein the central computer communicates with the separation.
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APPENDIX:

- Aebersold, R., B. Rist, et al. (2000). "Quantitative proteome analysis: methods and applications [In Process Citation]." Ann N Y Acad Sci 919: 33-47.
- Bennick, A. and F. Brosstad (1993). "A rapid method for selecting specific hybridoma clones using paramagnetic Dynabeads." Scand J Immunol 38(3): 212-4.
- Charlwood, J., J. M. Skehel, et al. (2000). "Analysis of N-linked oligosaccharides released from glycoproteins separated by two-dimensional gel electrophoresis." Anal Biochem 284(1): 49-59.
- Cordwell, S. J., A. S. Nouwens, et al. (2000). "Subproteomics based upon protein cellular location and relative solubilities in conjunction with composite two-dimensional electrophoresis gels." Electrophoresis 21(6): 1094-103.
- Corthals, G. L., V. C. Wasinger, et al. (2000). "The dynamic range of protein expression: a challenge for proteomic research." Electrophoresis 21(6): 1104-15.
- Dieden, R., R. K. Verbeeck, et al. (1999). "Isolation, identification and immunosuppressive activity of SDZ-IMM-125 metabolites from human liver microsomes." Eur J Drug Metab Pharmacokinet 24(1): 83-90.
- Gatlin, C. L., J. K. Eng, et al. (2000). "Automated identification of amino acid sequence variations in proteins by HPLC/microspray tandem mass spectrometry." Anal Chem 72(4): 757-63.
- Gebhardt, K., V. Lauvrak, et al. (1996). "Adhesive peptides selected by phage display: characterization, applications and similarities with fibrinogen." Pept Res 9(6): 269-78.
- Hayden, J. B., A. L. McCormack, et al. (1996). "Analysis of naturally processed peptides eluted from HLA DRB1*0402 and *0404." J Neurosci Res 45(6): 795-802.
- Islam, D. and A. A. Lindberg (1992). "Detection of *Shigella dysenteriae* type 1 and *Shigella flexneri* in feces by immunomagnetic isolation and polymerase chain reaction." J Clin Microbiol 30(11): 2801-6.
- Jackson, C. J., P. K. Garbett, et al. (1990). "Binding of human endothelium to *Ulex europaeus* I-coated Dynabeads: application to the isolation of microvascular endothelium." J Cell Sci 96(Pt 2): 257-62.

- Kandzia, J., W. Scholz, et al. (1984). "Magnetic albumin/protein A immunomicrospheres. I. Preparation, antibody binding capacity and chemical stability." J Immunol Methods 75(1): 31-41.
- 5 Kvalheim, G., O. Fodstad, et al. (1987). "Elimination of B-lymphoma cells from human bone marrow: model experiments using monodisperse magnetic particles coated with primary monoclonal antibodies." Cancer Res 47(3): 846-51.
- Link, A. J., J. Eng, et al. (1999). "Direct analysis of protein complexes using mass spectrometry." Nat Biotechnol 17(7): 676-82.
- 10 Meeusen, S., Q. Tieu, et al. (1999). "Mgm101p is a novel component of the mitochondrial nucleoid that binds DNA and is required for the repair of oxidatively damaged mitochondrial DNA." J Cell Biol 145(2): 291-304.
- Morel, V., R. Poschet, et al. (2000). "Towards the proteome of the rhodopsin-bearing post-Golgi compartment of retinal photoreceptor cells [In Process Citation]." Electrophoresis 21(16): 3460-9.
- 15 Owen, C. S. and J. G. Lindsay (1983). "Ferritin as a label for high-gradient magnetic separation." Biophys J 42(2): 145-50.
- Ozyhar, A., M. Gries, et al. (1992). "Magnetic DNA affinity purification of ecdysteroid receptor." J Steroid Biochem Mol Biol 43(7): 629-34.
- 20 Panigrahi, A. K., S. P. Gygi, et al. (2001). "Association of two novel proteins, TbMP52 and TbMP48, with the trypanosoma brucei RNA editing complex [In Process Citation]." Mol Cell Biol 21(2): 380-9.
- Saleh, A., D. Schieltz, et al. (1998). "Tra1p is a component of the yeast Ada.Spt transcriptional regulatory complexes." J Biol Chem 273(41): 26559-65.
- 25 Santoni, V., P. Dumas, et al. (1999). "Large scale characterization of plant plasma membrane proteins." Biochimie 81(6): 655-61.
- Santoni, V., T. Rabilloud, et al. (1999). "Towards the recovery of hydrophobic proteins on two-dimensional electrophoresis gels." Electrophoresis 20(4-5): 705-11.
- Scheffold, A., S. Miltenyi, et al. (1995). "Magnetofluorescent liposomes for increased sensitivity of immunofluorescence." Immunotechnology 1(2): 127-37.

- Simpson, R. J., L. M. Connolly, et al. (2000). "Proteomic analysis of the human colon carcinoma cell line (LIM 1215): development of a membrane protein database [In Process Citation]." Electrophoresis 21(9): 1707-32.
- Soskic, V., M. Gorlach, et al. (1999). "Functional proteomics analysis of signal transduction pathways of the platelet-derived growth factor beta receptor." Biochemistry 38(6): 1757-64.
- Spahr, C. S., S. A. Susin, et al. (2000). "Simplification of complex peptide mixtures for proteomic analysis: reversible biotinylation of cysteinyl peptides [In Process Citation]." Electrophoresis 21(9): 1635-50.
- Taylor, R. S., C. C. Wu, et al. (2000). "Proteomics of rat liver Golgi complex: minor proteins are identified through sequential fractionation [In Process Citation]." Electrophoresis 21(16): 3441-59.
- te Heesen, S., R. Rauhut, et al. (1991). "An essential 45 kDa yeast transmembrane protein reacts with anti- nuclear pore antibodies: purification of the protein, immunolocalization and cloning of the gene." Eur J Cell Biol 56(1): 8-18.
- Ushijima, H., H. Honma, et al. (1990). "Removal of HIV antigens and HIV-infected cells in vitro using immunomagnetic beads." J Virol Methods 29(1): 23-31.
- Yanagida, M., Y. Miura, et al. (2000). "Matrix assisted laser desorption/ionization-time of flight-mass spectrometry analysis of proteins detected by anti-phosphotyrosine antibody on two-dimensional-gels of fibroblast cell lysates after tumor necrosis factor-alpha stimulation [In Process Citation]." Electrophoresis 21(9): 1890-8.
- Yates, J. R., 3rd, E. Carmack, et al. (1999). "Automated protein identification using microcolumn liquid chromatography-tandem mass spectrometry." Methods Mol Biol 112: 553-69.
- Yates, J. R., 3rd, A. L. McCormack, et al. (1997). "Direct analysis of protein mixtures by tandem mass spectrometry." J Protein Chem 16(5): 495-7.
- Zhang, W., A. J. Czernik, et al. (1994). "Matrix-assisted laser desorption mass spectrometric peptide mapping of proteins separated by two-dimensional gel

electrophoresis: determination of phosphorylation in synapsin I." Protein Sci 3(4): 677-86.